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Product Sheet

IL-4 Reporter Cell Line

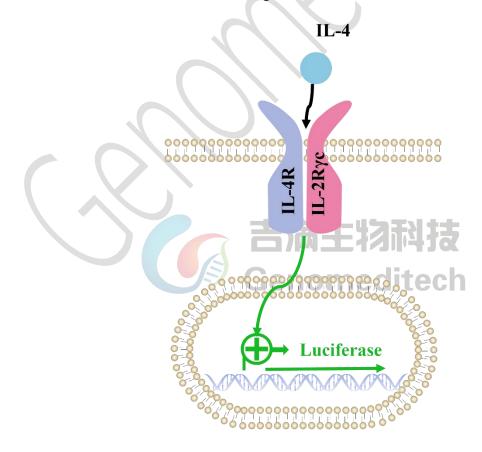
Catalog number: GM-C26301

Version 3.3.1.241121

Interleukin 4 (IL-4) is a cytokine produced by activated T cells, mast cells, and basophils, crucial for regulating immune responses and allergies. It promotes Th2-type responses, enhancing B cell proliferation and antibody production, particularly IgE and IgG1, while inhibiting Th1 responses to reduce inflammation.

IL-4 signals through its receptor IL-4R, type I receptor composed of IL-4R α and IL-2R γ chains, type II receptor composed of IL-4R α and IL-13R α 1 chains. Binding type I receptor activates JAK1 and JAK3, leading to STAT6 phosphorylation and nuclear translocation, triggering gene transcription. This pathway supports B cell growth, IgE production, and Th2 differentiation. IL-4 also influences cell growth via PI3K/Akt and MAPK pathways.

IL-4 Reporter Cell Line is a clonal stable cell line constructed using lentiviral technology, constitutive expression of the IL-4R α gene and IL-2R γ c gene, along with signal-dependent expression of a luciferase reporter gene. When IL-4 binds to IL-4R, it activates downstream signaling pathways, leading to the expression of luciferase. Blockade antibodies can inhibit this signal transmission. The luciferase activity measurement indicates the activation level of the signaling pathway and can thus be used to evaluate the in vitro effects of drugs related to IL-4.





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Specifications

Quantity 5E6 Cells per vial,1 mL

Product Format 1 vial of frozen cells

Shipping Shipped on dry ice

Storage Conditions Liquid nitrogen immediately upon receipt

Recovery Medium F12K+10% FBS+1% P.S

 $F12K+10\% \ FBS+1\% \ P.S+4 \ \mu g/mL \ Blasticidin+50 \ \mu g/mL \ Bleomycin+200 \ \mu g/mL \ G418+4$ Growth medium

μg/mL Puromycin

Note None

Freezing Medium 90% FBS+10% DMSO

Growth properties Adherent

Growth Conditions 37°C, 5% CO₂

Mycoplasma Testing The cell line has been screened to confirm the absence of Mycoplasma species.

Safety considerations Biosafety Level 2

Note It is recommended to expand the cell culture and store a minimum of 10 vials at an early

passage for potential future use.

Materials

Reagent	Manufacturer/Catalogue No.
F12K	BOSTER/PYG0036
Fetal Bovine Serum	Cegrogen biotech/A0500-3010
Pen/Strep	Thermo/15140-122
Blasticidin	Genomeditech/GM-040404
Bleomycin	Genomeditech/GM-040407
G418	Genomeditech/GM-040402
Puromycin	Genomeditech/GM-040401
Human IL4 / Interleukin-4 Protein	Sino Biological/GMP-11846-HNAE
Recombinant Human IL-13 Protein	Sino Biological/10369-HNAC
Anti-IL4R hIgG4 Antibody(Dupilumab)	Genomeditech/GM-53165AB
Anti-IL-4R hIgG1 Antibody(12B5)	Genomeditech/GM-46268AB
Anti-CD132(IL2RG) hIgG4 Antibody(REGN7257)	Genomeditech/GM-52334AB
PE anti-human CD213a1 (IL-13Rα1) Antibody	BioLegend/360403
GMOne-Step Luciferase Reporter Gene Assay Kit	Genomeditech/GM-040503

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Figures

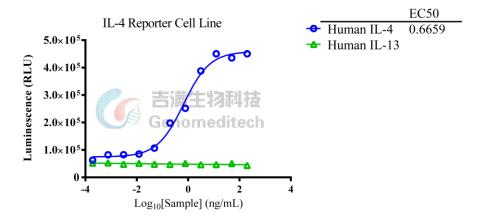


Figure 1 | Response to Human IL-4 Protein and Human IL13 Protein. IL-4 Reporter Cell Line (Cat. GM-C26301) at a concentration of 1.5E4 cells/well (96-well format) were stimulated with serial dilutions of Human IL4 Protein (SinoBiological/GMP-11846-HNAE) and Recombinant Human IL-13 Protein (SinoBiological/10369-HNAC) in assay buffer (F12K + 1% FBS + 1% P.S) for 7 hours. The firefly luciferase activity was measured using the GMOne-Step Luciferase Reporter Gene Assay Kit (Cat. GM-040503). The maximum induction fold of Human IL-4 Protein was approximately [10.6], while Human IL-13 protein, as expected, did not show any fold change. Data are shown by drug mass concentration.

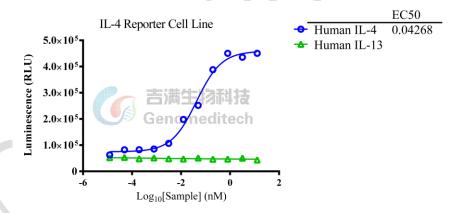


Figure 2 | Response to Human IL-4 Protein and Human IL13 Protein. IL-4 Reporter Cell Line (Cat. GM-C26301) at a concentration of 1.5E4 cells/well (96-well format) were stimulated with serial dilutions of Human IL4 Protein (SinoBiological/GMP-11846-HNAE) and Recombinant Human IL-13 Protein (SinoBiological/10369-HNAC) in assay buffer (F12K + 1% FBS + 1% P.S) for 7 hours. The firefly luciferase activity was measured using the GMOne-Step Luciferase Reporter Gene Assay Kit (Cat. GM-040503). The maximum induction fold of Human IL-4 Protein was approximately [10.6], and the validation of Human IL-13 Protein failed. Data are shown by drug molar concentration.



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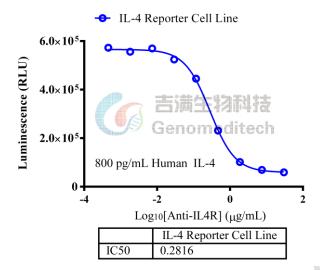


Figure 3 | Response to Anti-IL4R hIgG4 Antibody(Dupilumab). Serial dilutions of the Anti-IL4R hIgG4 Antibody(Dupilumab) (Cat. GM-53165AB) was incubated with 1.5E4 cells/well of the IL-4 Reporter Cell Line (Cat. GM-C26301) in a 96-well plate for 1 hour in assay buffer (F12K + 1% FBS + 1% P.S). Subsequently, the Human IL4 Protein (Sino Biological/GMP-11846-HNAE) at a final concentration of 800 pg/mL was added, and the coculture proceeded for an additional 6 hours. Firefly luciferase activity is then measured using the GMOne-Step Luciferase Reporter Gene Assay Kit (Cat.GM-040503). The results indicated maximum blocking folds of approximately [9.6]. Data are shown by drug mass concentration.

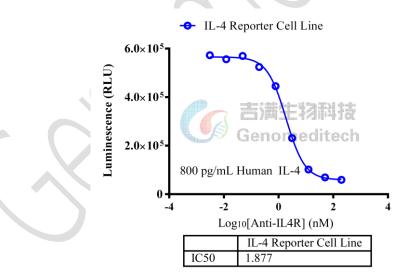


Figure 4 | Response to Anti-IL4R hIgG4 Antibody(Dupilumab). Serial dilutions of the Anti-IL4R hIgG4 Antibody(Dupilumab) (Cat. GM-53165AB) was incubated with 1.5E4 cells/well of the IL-4 Reporter Cell Line (Cat. GM-C26301) in a 96-well plate for 1 hour in assay buffer (F12K + 1% FBS + 1% P.S). Subsequently, the Human IL4 Protein (Sino Biological/GMP-11846-HNAE) at a final concentration of 800 pg/mL was added, and the coculture proceeded for an additional 6 hours. Firefly luciferase activity is then measured using the GMOne-Step Luciferase



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Reporter Gene Assay Kit (Cat.GM-040503). The results indicated maximum blocking folds of approximately [9.6]. Data are shown by drug molar concentration.

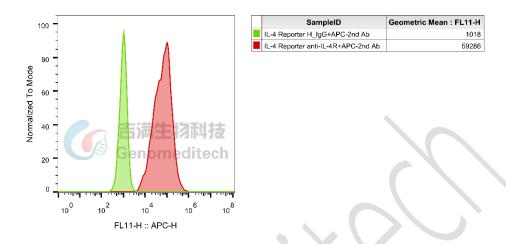


Figure 5 | IL-4 Reporter Cell Line (Cat. GM-C26301) was determined by flow cytometry using Anti-IL-4R hIgG1 Antibody(12B5) (Cat. GM-46268AB).

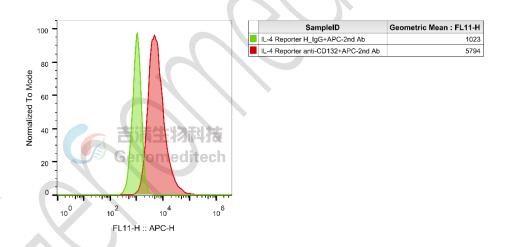


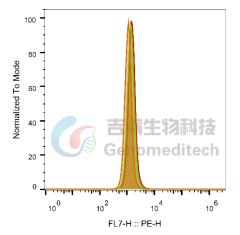
Figure 6 | IL-4 Reporter Cell Line (Cat. GM-C26301) was determined by flow cytometry using Anti-CD132(IL2RG) hIgG4 Antibody(REGN7257) (Cat. GM-52334AB).



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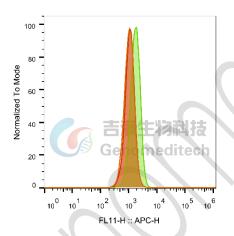
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SampleID	Geometric Mean : FL7-H
Null PE-anti-IL13RA1 Ab	1106
IL-4 Reporter PE-M_lgG Ab	1390
IL-4 Reporter PE-anti-IL13RA1 Ab	1514

Figure 7 | IL-4 Reporter Cell Line (Cat. GM-C26301) was determined by flow cytometry using APE anti-human CD213a1 (IL-13Ra1) Antibody (Biolegend/360403).



SampleID	Geometric Mean : FL11-H
Null anti-IL13RA2+APC-2nd Ab	1058
IL-4 Reporter H_IgG+APC-2nd Ab	1735
IL-4 Reporter anti-IL13RA2+APC-2nd Ab	1116

Figure 8 | IL-4 Reporter Cell Line (Cat. GM-C26301) was determined by flow cytometry using Anti-IL13RA2 hIgG1 Antibody (Genomeditech/In house).

Cell Recovery

Recovery Medium: F12K+10% FBS+1% P.S

To insure the highest level of viability, thaw the vial and initiate the culture as soon as possible upon receipt. If upon arrival, continued storage of the frozen culture is necessary, it should be stored in liquid nitrogen vapor phase and not at -70°C. Storage at -70°C will result in loss of viability.

Thaw the vial by gentle agitation in a 37°C water bath. To reduce the possibility of contamination, keep the O-ring and cap out of the water. Thawing should be rapid (approximately 2 - 3 minutes).



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- b) Remove the vial from the water bath as soon as the contents are thawed, and decontaminate by dipping in or spraying with 70% ethanol. All of the operations from this point on should be carried out under strict aseptic conditions.
- c) Transfer the vial contents to a centrifuge tube containing 5.0 mL complete culture medium and spin at approximately 176 x g for 5 minutes. Discard supernatant.
- d) Resuspend cell pellet with the recommended recovery medium. And dispense into appropriate culture dishes.
- e) Incubate the culture at 37°C in a suitable incubator. A 5% CO₂ in air atmosphere is recommended if using the medium described on this product sheet.

Cell Freezing

Freezing Medium: 90% FBS+10% DMSO

- a) Centrifuge at 176 x g for 3 minutes to collect cells.
- b) Resuspend the cells in pre-cooled freezing medium and adjust the cell density to 5E6 cells/mL.
- c) Aliquot 1 mL into each vial.
- d) Place the vial in a controlled-rate freezing container and store at -80°C for at least 1 day, then transfer to liquid nitrogen as soon as possible.

Cell passage

Growth medium: F12K+10% FBS+1% P.S+4 μ g/mL Blasticidin+50 μ g/mL Bleomycin+200 μ g/mL G418+4 μ g/mL Puromycin

For the first 1 to 2 passages post-resuscitation, use the recovery medium. Once the cells have stabilized, switch to a growth medium.

- a) Remove and discard culture medium.
- b) Briefly rinse the cell layer with PBS to remove all traces of serum that contains trypsin inhibitor.
- c) Add 1.0 mL of 0.25% (w/v) Trypsin-EDTA solution to dish and observe cells under an inverted microscope until cell layer is dispersed (usually within 2 to 3 minutes at 37°C).
- d) Note: To avoid clumping do not agitate the cells by hitting or shaking the flask while waiting for the cells to detach. Cells that are difficult to detach may be placed at 37°C to facilitate dispersal.
- e) Add 2.0 mL of growth medium to mix well and aspirate cells by gently pipetting.
- f) After centrifugation, resuspend the pellet and add appropriate aliquots of the cell suspension to new culture vessels.
- g) Incubate cultures at 37°C.

Subcultivation Ratio: A subcultivation ratio of 1:4 - 1:5 is recommended

Medium Renewal: Every 2 to 3 days

Notes

a) After the stabilization of the cell condition, there will be fewer dead cells post-passage, the cell growth rate will tend to stabilize, cell morphology will become uniform, and the cells will appear robust.



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Related Products

OX40		
H_OX40 Reporter Cell Line	Cynomolgus_OX40L CHO-K1 Cell Line	
H_OX40 CHO-K1 Cell Line	H_OX40L CHO-K1 Cell Line	
H_OX40L HEK-293 Cell Line		
Anti-H_OX40 hIgG2 Antibody(Ivuxolimab)	Anti-OX40L hIgG1 Reference Antibody(Oxebio)	
Anti-OX40L hIgG4 Antibody(Amlitelimab)	Anti-OX40L hIgG4 Reference Antibody(Amlbio)	
Biotinylated Human OX40L Protein; His-Avi Tag	Cynomolgus OX40 Protein; His Tag	
Cynomolgus OX40L Protein; His Tag	Cynomolgus OX40L Protein; mFc Tag	
Human OX40 Protein; His Tag	Human OX40L Protein; His Tag	
Human OX40L Protein; mFc Tag		
IL-4/IL-13		
IL-4/IL-13 Reporter 293 Cell Line	IL-4/IL-13 Reporter 293 DDX35TM Cell Line	
Cynomolgus_IL4R CHO-K1 Cell Line	H_IL4R CHO-K1 Cell Line	
Anti-IL-4R hIgG1 Antibody(12B5)	Anti-IL4R hIgG4 Antibody(Dupilumab)	
Anti-IL4R hIgG4 Reference Antibody (Dupbio)		
Human IL-4R alpha Protein; mFc Tag		
IL	-31	
H_IL-31 Reporter Cell Line	Cynomolgus_IL31RA CHO-K1 Cell Line	
H_IL31RA CHO-K1 Cell Line	H_IL31RA HEK-293 Cell Line	
H_IL-31RA OSMR Baf3 Cell Line		
Anti-IL31 hIgG1 Antibody(mAb33)	Anti-IL31RA hIgG1 Antibody(NA633)	
Anti-IL31RA hIgG2 Antibody(Nemolizumab)	Anti-OSMR hIgG4 Antibody(Vixarelimab)	
TSLP:TSLPR		
H_TSLP Reporter Cell Line	H_TSLPR CHO-K1 Cell Line	
Anti-H_TSLPR hIgG1 Antibody	Anti-TSLP hIgG2 Reference Antibody(Tezbio)	
Anti-TSLP hIgG2 Antibody(Tezepelumab)		
Cynomolgus TSLP Protein; His Tag	Human TSLP Protein; His Tag	
IL-5		
H_IL-5 Reporter 293 Cell Line	H_IL-5RA CHO-K1 Cell Line	
H_IL-5RA HEK-293 Cell Line		
Anti-IL5 hIgG4 Antibody(Reslizumab)	Anti-IL-5R hIgG1 Antibody(Benralizumab)	

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